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PROGRESSION OF SEXUAL EVOLUTION IN THE PLANT KINGDOM*

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Sexuality may manifest itself in at least three different ways: First, by the property of attraction and fusion of cells; second, by the dimorphism of characters expressed through the influence of the sexual state at the time of cell differentiation; and third, by a difference in the nature of the chemical bodies produced in cells in different sexual states. In the evolution of sex there are, apparently, degrees of intensity of the sexual state and degrees of persistency of any state established for the time, whether male, female, or neutral. The main factor in the evolution of sexual dimorphism has been the shifting of the time in the ontogeny when sexual states arise from the neutral state or when one sexual state gives place to another. The time of sex determination ranges all the way from a late stage of gametogenesis in the lowest forms backward through the ontogenetic history until, as in diecious flowering plants of the extreme type, the sexual state of both the sporophyte and gametophyte is established at the time of fertilization of the egg or even before. Of course, it must be remembered that new hereditary factors are continually arising or old ones being modified, in the upward progression of evolution, which come to expression either in the presence of one sexual state or the other or in the presence of both sexual states but with different morphological values. In either case the dimorphism of secondary sexual characters becomes more pronounced or more complex.

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If sexuality is a physiological state of the cells, then it becomes evident that sex reversal might take place at any time and in any cell or tissue. A neutral condition may change to a male state or to a female state, or one sexual state may give place to the other, or a sexual state may revert to a neutral state again. Such reversals not only take place naturally in hermaphroditic, bisporangiate, monecious and monosporangiate individuals, but they can be induced in various ways by artificial means in diecious sporophytes, as has been done by the writer and others in *Cannabis sativa*, *Humulus japonicus*, *Arisæma triphyllum*, and *Arisæma dracontium*.

If a distinction is to be made between different categories of sexual characters, then in order to avoid confusion of ideas and improper deductions, the terms "primary" and "secondary" need a more exact definition for application in genetics than they have received heretofore.

The proper place to draw the line is between gametes on the one hand and all other cells on the other. The ordinary dimorphic characters are produced because of the presence of a sexual state and not because of differences in heredity of the cells involved. A primary sexual character is, therefore, defined as any sexual character possessed by a gamete. A secondary sexual character is any sexual character possessed by any cell, tissue, or organ other than a gamete. Thus the wall cells and stalks of spermaries and ovaries may show secondary sexual characters comparable to those of cells farther away in the vegetative body. The main differences in primary sexual characters pertain to the size and shape of the gametes, size of or presence and absence of flagella, differences in activity, differences in food contents, and differences in color. The fundamental differences exhibited between eggs and sperms are often quite uniform throughout a class or phylum, while secondary sexual characters may be quite diverse.

The lowest plants are apparently devoid of sexual states during their entire life cycle, and the first indications of sex are entirely physiological. Passing on from this condition of a physiological state, the first appearance of a structural difference or dimorphism appears in the gametes. These differences soon reach a general, normal, dimorphic condition in the Gonidiophyta with well-differentiated eggs and sperms. So long as organisms are unicellular they cannot be described as her-

maphrodites. They must be either male or female although they may be vegetative sister cells. So soon as a multicellular condition is attained hermaphroditic individuals are possible, and in the lower multicellular forms hermaphroditism is the usual condition. If a progeny of unicellular individuals coming from an ancestral cell by vegetative growth develops some as male cells and some as female, we evidently have a condition essentially like an ordinary multicellular hermaphrodite; for if the cells remained associated after division instead of splitting apart the combined units going into the sexual state at a certain stage of their life history would represent genetical conditions identical to a multicellular hermaphrodite. The further evolution from hermaphrodites is by gradual steps to unisexual, multicellular individuals. In the higher plants with an antithetic alternation of generations, the sporophyte is in the lower levels entirely neutral and homosporous, showing no sexual states; and from this condition the higher bisporangiate type with heterospory is evolved. The heterosporous sporophytes of the lowest types have bisporangiate leaves or bisporangiate floral axes. This is the general condition from which both monecious and diecious species are derived. In the monecious individual the entire floral axis is monosporangiate or the entire inflorescence, while in the extreme diecious condition the entire individual produces but one kind of spores, or in other words has but one kind of sexual expression. The highest plants, therefore, have unisexual, dimorphic gametophytes and diecious, dimorphic sporophytes.

The detailed description of these evolutionary series is given in outline below although no attempt has been made to catalog the numerous intermediate and special cases.

I. NONSEXUAL STAGE.

1. The organization of the protoplast is apparently such that the origin of sexual states or phases is impossible. This may be true both morphologically and chemically. No sexual state is shown at any stage of the life cycle although there is considerable evolution and morphological differentiation displayed by the highest species of this type of plants.

Apparent examples: Merismopedia, Micrococcus, Ankistrodesmus, Rivularia.

II. LOWER SEXUAL SERIES, WITH REDUCTION OF THE CHROMOSOMES EITHER IN THE ZYGOTE, OR AT THE END OF THE SEXUAL GENERATION JUST BEFORE GAMETOGENESIS.

2. The lowest sexual condition in which sexuality manifests itself by the development of a passing state of attraction between two morphologically similar mature cells or isogametes. In the most primitive cases there is no apparent morphological or physiological differentiation outside of the property of attraction.

Examples: Diatoms, Desmids, Sphærella, Botrydium, Cladophora, Ulothrix, Ectocarpus.

Note: We may assume that there is a chemical or physical, or perhaps it would be better to say a physiological difference corresponding to female and male states.

3. Development of a difference in size and sometimes color of the gametes which in typical cases are ciliated, free-swimming cells.

Examples: Pandorina, Bryopsis, Codium, Spirogyra sp.

Note: Spirogyra shows a considerable degree of heterogamy in its highest species.

4. The stage of sexual evolution in which the gametes are typically differentiated as male and female with a decided dimorphism, the sperm cell being ciliated and small with little cytoplasm and food supply and actively motile after being discharged from the gametangium, the egg cell being nonciliated and comparatively large with abundant cytoplasm and food supply and also discharged from the gametangium although nonmotile.

Example: Fucus evanescens.

Note: Fucus also shows a dimorphism of the gametangia, and belongs to a more advanced stage of sexual evolution, but its female gametes represent a condition which is just a stage below the final evolutionary development of sexual cells.

5. The stage representing the attainment of typical female and male, plant gametes, showing the normal primary sexual dimorphism. Female gamete or egg—large, stationary (not discharged from the gametangium), with abundant cytoplasm and food supply; male gamete or sperm—small, free-swimming with cilia, and with a small amount of cytoplasm and food.

Examples: Volvox, Sphæroplea.

6. Hermaphroditic condition with a definite dimorphism showing in the gametangia but not in the contiguous regions or only to a very slight extent. In these plants the sex is determined in the incipient gametangia.

Examples: Monoblepharis, Vaucheria sessilis, Coleochæte.

7. Hermaphrodites having a greater or less area of tissue immediately beyond the gametangia proper showing sexual dimorphism. The sexual state is determined in the tissues before the definite origin of the gametangia and so influences the morphological expression in the tissues below them.

Examples: Chara, Nitella.

8. Hermaphrodites having considerable areas of their bodies differentiated as male or female tissues or branches from which only spermaries or ovaries are produced. The sex is determined in the vegetative tissue or branch long before gametangia are developed.

Examples: Vaucheria synandra, Oedocladium protonema. Fucus evanescens.

9. The culmination stage in the series represented by unisexual individuals of various degrees of intensity and stability of the male or female state and showing some dimorphism in the entire individual. The sexual state in such individuals is determined either in the gametes before fertilization, in the zygote, or in a very early stage after germination of the zygote. There are probably two progressive phases, which, however, can usually be determined only by experiment: (*a*) Species in which sexuality is not strongly fixed, that is the hereditary constitution is such that a reversal of the sexual state is readily brought about in the individual. (*b*) Species in which the sexual state, either male or female, seems to be strongly fixed in the individual, so that it becomes difficult to change from one sexual state to a neutral state or from one sexual state to the other.

Examples: Oedogonium lautumniarum, Fucus vesiculosus, Mucor stolonifer (specialized sexual condition).

III. HOMOSPOROUS SERIES WITH ANTITHETIC ALTERNATION OF GENERATIONS.

In this series the sporophyte shows no sexual differentiation whatever, but in some species there may possibly be a physiological difference in the spores with some cytological dimor-

phism, as a chromosome difference, for example. The sporophyte is nonsexual in expression in every respect although it carries sexual potentialities in its cells in a latent condition. These potentialities become evident in such abnormal cases as apospory. The sporophyte in normal conditions is entirely neutral in respect to sex, while the gametophyte shows all gradations from hermaphrodites with closely associated sex organs to extreme, unisexual individuals.

10. The lowest stage of this series is represented by species with haploid, hermaphroditic gametophytes having the ovaries and spermaries closely associated. Examples: many liverworts and mosses (synoicous condition).

11. Haploid gametophytes which are hermaphrodites, but which have distinctly differentiated male and female areas. Examples: many ferns, mosses, and liverworts as Lophocolea heterophylla (paroicous condition).

12. Haploid hermaphroditic gametophytes which have the male and female organs on distinct branches or axes. Examples: many liverworts, bog-mosses, and mosses, as Phascum cuspidatum (autoicous condition).

There are, of course, practically all possible intergradations between stages 10, 11 and 12.

13. Haploid gametophytes differentiated into male and female individuals (unisexual) but which may be reversed in sex, either normally through age and growth or artificially by being subjected to the proper environment.

Examples: Equisetum arvense, Matteuccia struthiopteris.

Note: There are also many species especially among the liverworts and mosses which have part of the individuals unisexual and part hermaphroditic (imperfectly unisexual), as Cephalozia curvifolia.

14. Haploid gametophytes differentiated into male and female individuals and these apparently not reversible by known experimental means or reversible only rarely; often showing a high degree of sexual dimorphism; in a few cases at least, but apparently not often, with a difference in size of synaptic chromosomes which are associated with the two sexes.

Examples: Sphærocarpus, Marchantia, Polytrichum.

Note: Marchantia polymorpha is occasionally hermaphroditic.

IV. HETEROSPOROUS SERIES WITH ANTITHETIC ALTERNATION OF GENERATIONS, THE GAMETOPHYTES BEING UNISEXUAL AND THE SPOROPHYTE INDIVIDUALS BISPORANGIATE OR MONOSPORANGIATE.

In this series the sporophyte shows dimorphism at least in the spores and sporangia. The gametophyte generation is apparently always unisexual with distinct male and female individuals highly differentiated both in morphological and physiological characteristics. Normally, the sexual state of the gametophytes is irreversible and determined even before the spores are produced from which they originate.

15. The lowest stage of this series is represented by plants whose bisporangiate sporophytes have only a slight dimorphism; only the spores and sporangia are differentiated and show male and female expression. The two kinds of sporangia are developed side by side on the same leaflet and sorus. The sexual expression is controllable to a slight extent by artificial means.

Example: Marsilea.

Note: It must be remembered that this condition is evolving from a sporophyte that shows absolutely no sexuality in morphological expression in any part of its body but that its cells are, nevertheless, potentially sexual, the gametophytes possessing a high degree of sexual dimorphism.

16. The first advance on the preceding condition is slight but important. This stage is represented by bisporangiate sporophytes with distinct microsporangiate and megasporangiate sori which develop into sporocarps but on the same leaf and which may be side by side. The sexual state is determined considerably earlier than the incepts of the sporangia in the group of cells or in the tissues from which the entire sorus is developed. The given sexual state must, however, be rather weak at first since vestiges of the opposite organs appear.

Examples: Azolla caroliniana and Salvinia natans.

Note: The megasporangiate sorus of Azolla contains vestigial microsporangia and the microsporangiate sorus contains a vestigial megasporangium, plainly showing that the monosporangiate condition of the sorus was derived from an original bisporangiate sorus as in Marsilea, the beginning of the sexual state being thrown back to some extent. The change from one sexual state to the other in the incipent sporangia inhibits the full development.

17. Bisporangiate sporophytes with the entire sporophylls more or less differentiated and with but one kind of sporangia, but the sporophylls arising side by side from a common stem tissue. The typical state of the higher plants.

Examples: Selaginella, Bennettitites, Magnolia, Aquilegia, Lilium, Cypripedium, Lactuca.

18. Monecious sporophytes in which the entire floral axis with its parts is differentiated, the staminate and carpellate flowers commingled more or less closely and showing large vestiges of the opposite set of organs.

Examples: Cocos, Aesculus.

Note: The suppression of the opposite set of sporophylls is of every degree, from this stage on, and has no relation to the area of tissue involved in the sexual differentiation. There are species in which the axis of the inflorescence is at first neutral and gives rise to bisporangiate flowers below and then changes above to a condition leading to the male state when only staminate flowers are produced.

Example: Lophotocarpus calycinus. Such species have an intermediate position between stages 17 and 18.

19. Monecious sporophytes having the axis of the inflorescence differentiated, the staminate flowers being developed first and, by a reversal of the sexual state, the carpellate later, or more commonly the carpellate flowers first and the staminate later.

Examples: Cymophyllus fraseri, Carex nardina, Tripsacum dactyloides, Zizania aquatica, Ricinus communis, Musa sapientum, Stillingia sylvatica. Typha latifolia.

In such inflorescences bisporangiate or abnormal flowers frequently appear on the neutral transition zone where the sexual state changes from one condition to the other.

20. Monecious sporophytes having an entire branch or inflorescence determined as staminate or carpellate.

Examples: Carex lupulina, Euchlæna mexicana, Zea mays (normal form).

21. The lower type of diecious sporophytes having only a moderate reduction in the size of the opposite sporophylls in the monosporangiate flowers, with frequent reversals to the opposite sexual condition—staminate plants developing carpellate flowers and carpellate plants staminate flowers.

Examples: Acer platanoides, Acer rubrum.

There are, of course, interesting intermediate developments, as for example one type of individuals may be monecious and the other purely staminate. An example of this condition is Arisæma dracontium.

22. Diecious sporophytes with complete or nearly complete suppression of the opposite set of organs, in normal cases in all the flowers of an individual, and having a more or less decided vegetative dimorphism. These diecious sporophytes often have a considerable percentage of intermediate individuals or are known by experiment to show frequent reversals in sexual expression, either through mutations, or through the influence of the environment.

Examples: Arisæma triphyllum, Morus alba, Morus rubra, Cannabis sativa, Humulus japonicus, Mercurialis annua.

23. The last and most extreme type with diecious sporophytes apparently without intermediates and not readily or not at all reversible in sexual expression. When the sexual state is once established, the staminate plants remain pure staminate and the carpellate pure carpellate.

Examples: Acer negundo, Populus deltoides, Fraximus americana.

Note: As stated, by such species is represented the extreme limit of evolution in relation to sexual expression. The gametophyte consists of pure male and female individuals apparently irreversible in the normal environment, and the sporophyte consists of pure staminate and carpellate individuals whose sexual state is apparently fixed in their normal environmental conditions. It is probable, however, that all diecious sporophytes will be found reversible as to sex under the proper environment during development. In these plants both the gametophytes and sporophytes are dimorphic. The sexual state of the gametophyte is determined in the previous sporophyte generation and the sexual state of the sporophyte is determined either in the gametes before fertilization, in the zygote at the time of fertilization, or in the embryo after the germination of the zygote.

DISCUSSION OF THE FOREGOING SERIES.

From a study of the preceding series of species and their numerous near relatives, from the lowest manifestation of sexuality to the highest, it becomes evident that sexuality in respect to its influence on the morphology of the vegetative structures has been a gradual evolution making a slow and almost uniform progress to the ultimate species, while the evolution of the gametes, although also showing fundamentally a similar progression, was attained very early, so that usually, when one passes a little beyond the unicellular forms and simple filaments, one finds the same general differences between eggs and sperms as are present in the highest types. The most remarkable thing about the whole evolutionary progression is the fact that very rarely does any step in advancement have any direct relation either to the reduction or to the fertilization stage. So it comes about that the determination of the sexual state takes place in the vast majority of cases in the vegetative tissues, at other times than the periods of reduction and fertilization. On the other hand, since hereditary factors are apparently properties of chromosomes, the characters of the individual outside of its sexual state are in the vast majority of cases predetermined by reduction and fertilization. The environment merely determines the degree of hereditary expression or the time of the expression in the life history.

We can then postulate the phenomena of sexuality as distinct from hereditary units. Probably ordinary hereditary units are irreversible properties of specific parts of the chromatin and cannot be changed except by mutation, while sexual states are due to fundamental properties of the protoplasm or its secretions due to a chemical or physical condition of the molecules or atoms and so any given organic structure may pass from the female to the neutral and then to the male state, or in any direction from one of these states to the other without any change in the hereditary factors whatever.

Cells may, therefore, be absolutely nonsexual or potentially sexual. The potentially sexual may be in the male, female, or neutral state in varying degrees of intensity. It has been the practice of morphologists to designate individuals and generations in an antithetic cycle as "nonsexual" and there is no special objection to this although "neutral" individual or gen-

eration might perhaps define the condition somewhat better, but objections might also be raised to this term as applied to heterosporous sporophytes. If we designate individuals by the kind of reproductive cells they produce, then there is no confusion in designating the sporophyte as "nonsexual." It is probably best at present to hold that the lowest forms are entirely nonsexual and not merely neutral, because their protoplasts apparently do not have the proper complexity of organization which would permit of or lead to sexual states.

The first differentiation then, as shown above, is in the gametes themselves, and apparently the most fundamental manifestation of sexuality is one of reaction or attraction of two cells (gametes) toward each other. If the sexual state arises in a cell, destined to be a gamete, after its morphological expression is practically completed then there can be no structural dimorphism, but the earlier the maleness or femaleness is established in the cell the more extreme the dimorphism must become. Since there can be only a short transition between the two extremes of isogamy and heterogamy the normal heterogamous state is soon attained, often with little or no advance in the evolution of vegetative parts, as stated before. The typical egg as has long been known, is comparatively large with a large amount of cytoplasm and food content and is nonmotile, lacking cillia and flagella, while the sperm is small and motile, being provided with cilia or flagella. These distinctions are true for the great majority of types of plants and animals, from the lowest heterogamous species to the highest. The greatest deviation is in respect to the motile organs of the sperm, which are absent in the rhodophyta, strobilophyta, and anthophyta. In other respects, however, the typical dimorphism is not materially altered. Secondary sexual characters begin to appear when a given sexual state is established in the neutral vegetative cells whose descendants are not all transformed into gametes or when a given sexual state is established in cells which are destined to produce vegetative structures only or at least before the final gamete-producing tissue is developed. Finally the dimorphism may involve the entire individual. In this case the sexual state must be present in the spore from which the individual develops or soon after its germination. The conditions which determine whether a cell or tissue shall pass into the male or female state are probably diverse, but

favorable and unfavorable nutritive conditions and maturity or senility seem to be among the most important factors. Disturbances brought about by unequal distribution of autosomes or characteristic distributions of allosomes appear also to be conditions bringing about or influencing sexual states, probably by an influence on the metabolism of the cell. Besides these cases in which sexual states are associated with slight differences in chromosome numbers are those in which a haploid or diploid set may be associated with one sex or the other. It is well-known, however, that doubling the number of chromosomes in a gametophyte does not necessarily disturb the sexual condition. It would appear that in cells with allosomes sex reversal should be more difficult to bring about than in cells which have equal distributions of chromosomes, but this is for future experiments to determine. It is well known that in numerous cases secondary sexual characters are reversed in spite of the presence of allosomes. The probable reason that secondary sexual characters become more prominent in the higher forms is because the protoplast contains a greater number of hereditary factors which can be influenced by the sexual state at the time of their expression of characters.

It becomes evident that sex cannot be associated primarily with special chromosomes. The hypothesis of homozygous or heterozygous combinations as determiners and controllers of any given sexual state falls entirely outside of the complex phenomena to be explained. Neither hereditary factors for sex nor factors which control sex are in evidence; for in the vast majority of plants the sexual state is determined in the somatic cells in which neither segregation nor association of chromosomes is taking place. So far as the gametophyte is concerned, after passing through the evolutionary phase where hermaphrodites are the rule, it is illuminating in the highest degree to find that when the complete segregation of sex was attained in the gametophyte it was not at all by the separation of units with sexual control in the reduction division, but at a different stage of the life cycle, namely, in the vegetative cells of the sporophyte. Thus the megasporocytes do not segregate maleness from femaleness nor any properties or hereditary factors which exercise a control over sexual expression, but all the cells of a megasporocyte are differentiated with femaleness, and give rise to female gametophytes, and in the same way the

microsporocytes give rise to four microspores, as the result of reduction, and all of the four develop into male gametophytes. The reduction division in this case as in others segregates the chromosomes with their complements of Mendelian factors, but sexuality is not involved in such segregation and is not affected or changed by it. It is, therefore, self evident that fundamental sexual phenomena are caused by properties in plants entirely apart from Mendelian units.

The specific structures and functions developed in the ontogeny of an organism appear to be conditioned on the interaction of four fundamental influences: (1) the hereditary factors themselves, apparently properties of the chromosomes; (2) the influence of the environment, both external and internal; (3) the progression of senility, probably including a fundamental exhaustion and degeneration of the protoplast as well as retardation or change of its activities due to chemical deposits; and (4) the presence of sexual states in the living substance, through which primary and secondary sexual characters and functions are produced and which are probably positive and negative states of atoms or molecules contained in the living cell.